

AMENDMENTS TO CLAIMS

Amendments to the claims are reflected in the following listing of claims, which replaces all prior versions or listings of the claims.

1 -46. (Canceled)

47. (Previously presented) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with a coding sequence, derived from *Bacillus*, that encodes an insecticidal polypeptide and that contains a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table II;

(b) reducing the number of said ATTTA sequences or the number of said polyadenylation signal sequences in the coding sequence by substituting sense codons for codons in the coding sequence; and

(c) making a structural gene that comprises a coding sequence that includes the codons substituted according to step (b) and is characterized by the reduced number of ATTTA sequences or Table II polyadenylation signal sequences, and that encodes an insecticidal protein.

48. (Previously presented) The method of claim 47, wherein step (b) comprises reducing the number of said polyadenylation signal sequences in the coding sequence.

49. (Previously presented) The method of claim 47, wherein step (b) comprises reducing the number of said ATTTA sequences in the coding sequence.

50. (Previously presented) The method of claim 47, wherein step (b) comprises reducing the number of said ATTTA sequences and the number of said polyadenylation signal sequences in the coding sequence.

51. (Previously presented) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with a coding sequence, derived from *Bacillus*, that encodes an insecticidal polypeptide and that contains a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table II;

(b) reducing the number of said ATTTA sequences or the number of said polyadenylation signal sequences in a portion of the coding sequence by substituting sense codons for codons in said portion; and

(c) making a structural gene that comprises said portion with the substitute codons and the reduced number of ATTTA or polyadenylation signal sequences, wherein the structural gene comprises a nucleotide sequence that encodes an insecticidal protein.

52. (Previously presented) The method of claim 51, wherein step (b) comprises reducing the number of said ATTTA sequences in said portion.

53. (Previously presented) The method of claim 51, wherein step (b) comprises reducing the number of said polyadenylation signal sequences in said portion.

54. (Previously presented) The method of claim 51, wherein step (b) comprises reducing the number of said ATTTA sequences and the number of said polyadenylation signal sequences in said portion.

55. (Currently amended) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with coding sequences, from one or more structural genes derived from *Bacillus*, that encode an insecticidal polypeptide and that ~~contains~~ contain a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table II;

(b) reducing the number of said ATTTA sequences or the number of said polyadenylation signal sequences in the coding sequences by substituting sense codons for codons in the coding sequences; and

(c) making a structural gene that comprises the coding sequences with the codons substituted according to step (b) and characterized by the reduced number of ATTTA or polyadenylation signal sequences, wherein the structural gene comprises a nucleotide sequence that encodes an insecticidal protein.

56. (Previously presented) The method of claim 55, wherein step (b) comprises reducing the number of said ATTTA sequences in said coding sequences.

57. (Previously presented) The method of claim 55, wherein step (b) comprises reducing the number of said polyadenylation signal sequences in said coding sequences.

58. (Previously presented) The method of claim 55, wherein step (b) comprises reducing the number of said ATTTA sequences and the number of said polyadenylation signal sequences in said coding sequences.

59. (Previously presented) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with an amino acid sequence of an insecticidal protein derived from *Bacillus*, wherein wild-type *Bacillus* gene sequence(s) encoding insecticidal polypeptide(s) from which the insecticidal protein is derived comprise a plurality of nucleotide sequences selected from the group consisting of ATTTA sequence(s) and polyadenylation signal sequences listed in Table II; and

(b) making a structural gene that comprises a coding sequence that:

(i) encodes the amino acid sequence of the insecticidal protein; and

(ii) contains fewer polyadenylation signal sequences listed in Table II or fewer ATTTA sequences, compared to the corresponding coding sequence(s) of the wild-type *Bacillus* gene sequences(s).

60. (Previously presented) The method of claim 59, wherein the wild-type *Bacillus* gene sequence(s) comprise a plurality of ATTTA sequences, and wherein the structural gene made according to step (b) contains fewer ATTTA sequences compared to the wild-type *Bacillus* gene sequences(s).

61. (Previously presented) The method of claim 59, wherein the wild-type *Bacillus* gene sequence(s) comprise a plurality of said polyadenylation signal sequences, and wherein the structural gene made according to step (b) contains fewer of said polyadenylation signal sequences compared to the wild-type *Bacillus* gene sequence(s).

62. (Previously presented) The method of claim 59, wherein the wild-type *Bacillus* gene sequence(s) comprise a plurality of both said ATTTA and said polyadenylation signal sequences, and wherein the structural gene made according to step (b) contains fewer

ATTTA sequences and fewer polyadenylation signal sequences compared to the wild-type *Bacillus* gene sequence(s).

63. (Currently amended) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with an amino acid sequence of an insecticidal protein derived from *Bacillus*; and

(b) making a structural gene that comprises a coding sequence that encodes the amino acid sequence and that is devoid or substantially devoid of ATTTA sequences, or devoid or substantially devoid of polyadenylation signal sequences listed in Table II.

64. (Currently amended) The method of claim 63, wherein step (b) comprises making a structural gene that is devoid or substantially devoid of the ATTTA sequences.

65. (Currently amended) The method of claim 63, wherein step (b) comprises making a structural gene that comprises a coding sequence that is devoid or substantially devoid of the polyadenylation signal sequences.

66. (Currently amended) The method of claim 63, wherein step (b) comprises making a structural gene that comprises a coding sequence that is devoid or substantially devoid of the ATTTA sequences and devoid or substantially devoid of the polyadenylation signal sequences.

67. (Currently amended) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with coding sequences that encode portions of one or more insecticidal polypeptides derived from *Bacillus*; and

(b) combining the coding sequences to form a structural gene that encodes an insecticidal protein,

wherein said coding sequences and the structural gene are devoid or substantially devoid of polyadenylation signal sequences listed in Table II.

68. (Currently amended) The method of claim 67, wherein said coding sequences and the structural gene are devoid or substantially devoid of ATTTA sequences.

69. (Previously presented) The method according to any one of claims 47, 51, and 55, wherein the structural gene made according to the method is more highly expressed in a dicot plant cell than a structural gene that comprises the starting coding sequence(s) of step (a).

70. (Previously presented) The method according to any one of claims 59 and 67, wherein the structural gene made according to the method is more highly expressed in a dicot plant cell than a structural gene that comprises the wild-type *Bacillus* gene sequences encoding polypeptide(s) from which the amino acid sequence of the insecticidal protein is derived.

71. (Previously presented) The method according to any one of claims 49, 52, 56, and 60, wherein the structural gene made according to the method contains no more than seven ATTTA sequences.

72. (Previously presented) The method according to any one of claims 49, 52, 56, 60, 64, and 68, wherein the structural gene made according to the method contains no greater than one ATTTA sequence.

73. (Previously presented) The method according to any one of claims 49, 52, 56, 60, 64, and 68, wherein the structural gene made according to the method contains no ATTTA sequences.

74. (Previously presented) The method according to any one of claims 48, 53, 57, and 61, wherein the structural gene made according to the method contains no more than seven of said polyadenylation signal sequences.

75. (Previously presented) The method according to any one of claims 48, 53, 57, 61, and 67, wherein the structural gene made according to the method contains no more than two of said polyadenylation signal sequences listed in Table II.

76. (Previously presented) The method according to any one of claims 48, 53, 57, 61, 63, and 67, wherein the structural gene made according to the method contains no greater than one of said polyadenylation signal sequences.

77. (Previously presented) The method according to any one of claims 48, 53, 57, 61, 63, and 67, wherein the structural gene made according to the method contains no polyadenylation signal sequences listed in Table II.

78. (Previously presented) The method according to any one of claims 47, 51, 55, 59, 63, and 67, wherein the structural gene made according to the method contains a (G+C) content of about 50%.

79. (Previously presented) The method according to any one of claims 47, 51, 55 and 67, wherein the starting coding sequence(s) of step (a) has (have) an (A + T) content of about 62%.

80. (Previously presented) The method according to any one of claims 59 and 63, wherein the wild-type *Bacillus* gene sequences have an (A+ T) content of about 62%.

81. (Previously presented) The method according to any one of claims 47, 51, 55, and 67, wherein the starting coding sequence(s) of step (a) are derived from *Bacillus thuringiensis* (*B.t.*).

82. (Previously presented) The method according to any one of claims 47, 51, 55 and 67, wherein the starting coding sequence(s) of step (a) are derived from a *Bacillus thuringiensis* (*B.t.*) crystal protein gene.

83. (Previously presented) The method of any one of claims 47, 51, 55, and 67, wherein the starting coding sequence(s) of step (a) are derived from a *Bacillus thuringiensis* (*B.t.*) P2 protein or a *B.t. entomocidus* gene.

84. (Previously presented) The method according to any one of claims 59 and 62, wherein the wild-type gene sequences are from *Bacillus thuringiensis* (*B.t.*).

85. (Previously presented) The method according to any one of claims 59 and 62, wherein the wild-type gene sequences comprise *Bacillus thuringiensis* (*B.t.*) crystal protein gene sequences.

86. (Previously presented) The method according to any one of claims 59 and 62, wherein the wild-type gene sequences comprise *Bacillus thuringiensis* (*B.t.*) P2 gene sequences or *B.t. entomocidus* gene sequences.

87. (Previously presented) The method according to any one of claims 63 and 66-68, wherein the insecticidal protein is derived from *Bacillus thuringiensis* (*B.t.*).

88. (Previously presented) The method according to any one of claims 47, 51, 59, 63, and 67, wherein the insecticidal protein is a *Bacillus thuringiensis* (*B.t.*) crystal protein.

89. (Previously presented) The method according to any of claims 47, 51, 55 and 67, wherein the starting coding sequence(s) of step (a) are derived from *B.t. tenebrionus*.

90. (Previously presented) The method according to any one of claims 47, 51, 59, and 63, wherein the insecticidal protein is a *B.t.* P2 protein or a *B.t. entomocidus* protein.

91. (Previously presented) The method according to any one of claims 47, 51, and 55, wherein the coding sequence(s) of step (a) comprise(s) a hybrid of coding sequences of at least two insecticidal proteins from *B.t.*

92. (Previously presented) The method according to any one of claims 59, 63, and 67, wherein the insecticidal protein comprises a hybrid of *B.t.* insecticidal proteins.

93. (Previously presented) The method according to any one of claims 47, 51, 55, and 67, wherein the coding sequence(s) of step (a) encode(s) an insecticidal fragment of a *Bacillus* insecticidal protein.

94. (Previously presented) The method according to any one of claims 47, 51, 55, and 67, wherein the coding sequence(s) of step (a) encode(s) a full length *Bacillus* insecticidal protein.

95. (Previously presented) The method according to any one of claims 59 and 63, wherein the insecticidal protein derived from *Bacillus* comprises an insecticidal fragment of a *Bacillus* insecticidal protein.

96. (Previously presented) The method according to any one of claims 59 and 63, wherein the insecticidal protein derived from *Bacillus* comprises a full length *Bacillus* insecticidal protein.

97. (Previously presented) The method according to any one of claims 47, 51, and 55, wherein the substituting of sense codons does not change the amino acid sequence encoded by the coding sequence.

98. (Previously presented) The method according to any one of claims 47, 51, 55, 59, 63, and 67, wherein the insecticidal protein encoded by the structural gene comprises an amino acid sequence that is identical to the amino acid sequence of an insecticidal protein from *Bacillus*, or an insecticidal fragment thereof.

99. (Previously presented) The method according to any one of claims 47, 51, 55, 59, and 67, wherein the insecticidal protein and the insecticidal polypeptide have the same amino acid sequence.

100. (Previously presented) The method according to any one of claims 47, 51, 55, 59, and 63, comprising avoiding the introduction of sense codons that are rarely found in plant genomes into the resultant structural gene.

101. (Previously presented) The method according to any one of claims 47, 51, 55, 59, and 63, comprising avoiding, in the resultant structural gene, the introduction of sense codons that contain a TA doublet.

102. (Previously presented) The method according to any one of claims 47, 51, 55, 59, and 63, comprising avoiding, in the resultant structural gene, the introduction of sense codons that contain a CG doublet.

103. (Previously presented) The method according to any one of claims 47 and 55, further comprising reducing the number of regions in the coding sequence(s) with greater than five consecutive adenine and thymine (A+T) nucleotides by substituting sense codons for codons in the coding sequence(s).

104. (Previously presented) The method according to claim 51, further comprising reducing the number of regions in said portion with greater than five consecutive adenine and thymine (A+T) nucleotides by substituting sense codons for codons in the portion.

105. (Previously presented) The method according to any one of claims 59, 63, and 67, wherein the structural gene comprises a coding sequence that does not contain more than five consecutive adenine and thymine (A+T) nucleotides.

106. (Previously presented) The method according to any one of claims 47 and 55, further comprising truncating the coding sequence to yield a truncated structural gene that encodes a truncated protein that retains insecticidal activity.

107. (Previously presented) The method according to any one of claims 47, 51, 55, 59, 63, and 67, further comprising attaching a plant promoter to the structural gene.

108. (Previously presented) The method according to any one of claims 47, 51, 55, 59, 63, and 67, further comprising including in the structural gene a sequence that encodes an amino-terminal chloroplast transit peptide or a secretory signal sequence.

109. (Previously presented) The method according to any one of claims 47, 51, 55, 59, 63, and 67, further comprising attaching to the structural gene a 3' non-translated nucleotide sequence that comprises a plant polyadenylation signal.

110. (Previously presented) The method according to any one of claims 47, 51, and 55, wherein the making step comprises performing site directed mutagenesis on a coding sequence from *Bacillus* to make the structural gene.

111. (Previously presented) The method according to any one of claims 47, 51, and 55, wherein the making comprises *de novo* synthesis of a fully synthetic structural gene.

112. (Currently amended) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with a sequence derived from *Bacillus*, said sequence comprising a coding sequence for an insecticidal protein, or an amino acid sequence of the insecticidal protein; and

(b) making a structural gene that comprises a coding sequence that encodes the amino acid sequence and that is devoid or substantially devoid of ATTTA sequences, or devoid or substantially devoid of polyadenylation signal sequences listed in Table II.

113. (Previously presented) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) designing a nucleotide sequence that encodes an insecticidal protein derived from *Bacillus* and that contains a reduced number of ATTTA sequences or Table II polyadenylation signal sequences, compared to wild type *Bacillus* coding sequence(s) from which the insecticidal protein was derived; and

(b) making a structural gene that comprises the nucleotide sequence, that encodes the insecticidal protein, and that is characterized by the reduced number of ATTTA sequences or Table II polyadenylation signal sequences, compared to the wild type *Bacillus* coding sequence(s).

114. (Previously presented) The method of claim 113, wherein the insecticidal protein comprises a protein selected from the group consisting of: (a) *Bacillus thuringiensis* (*B.t.*) insecticidal proteins; (b) insecticidal fragments of (a); and insecticidal fusions derived from more than one *B.t.* insecticidal protein.

115. (Previously presented) The method of claim 113, wherein the wild-type *Bacillus* coding sequence(s) comprise a plurality of both said ATTTA and said polyadenylation signal sequences, and wherein the structural gene made according to step (b) contains fewer ATTTA sequences and fewer polyadenylation signal sequences compared to the wild-type *Bacillus* coding sequence(s).

116. (Previously presented) The method according to claim 113, wherein the structural gene made according to the method is more highly expressed in a dicot plant cell than a structural gene that consists of wild type *Bacillus* coding sequence(s) from which the insecticidal protein was derived.

117. (Currently amended) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) designing a nucleotide sequence that encodes an insecticidal protein derived from *Bacillus* and that is devoid or substantially devoid of ATTTA sequences, or devoid or substantially devoid of Table II polyadenylation signal sequences; and

(b) making a structural gene that comprises the nucleotide sequence and that encodes the insecticidal protein, wherein the structural gene is devoid or substantially devoid of ATTTA sequences, or devoid or substantially devoid of Table II polyadenylation signal sequences.

118. (Currently amended) The method of claim 117, wherein the structural gene is devoid or substantially devoid of the ATTTA sequences, and devoid or substantially devoid of the polyadenylation signal sequences.

119. (Previously presented) A method of making a structural gene that encodes an insecticidal protein, the method comprising:

(a) starting with a coding sequence, derived from *Bacillus*, that contains a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table I, and that encodes an insecticidal protein having an amino acid sequence;

(b) making a structural gene that comprises a coding sequence that:

(i) encodes the amino acid sequence of the insecticidal protein; and

(ii) contains fewer polyadenylation signal sequences listed in Table II or fewer ATTTA sequences, compared to the corresponding coding sequence derived from *Bacillus*.

120. (New) A method of making a structural gene that encodes a protein, the method comprising:

(a) starting with a coding sequence that encodes a polypeptide and that contains a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table II;

(b) reducing the number of said ATTTA sequences or the number of said polyadenylation signal sequences in the coding sequence by substituting sense codons for codons in the coding sequence; and

(c) making a structural gene that comprises a coding sequence that includes the codons substituted according to step (b) and is characterized by the reduced number of ATTTA sequences or Table II polyadenylation signal sequences, and that encodes a protein.

121. (New) The method of claim 120, wherein step (b) comprises reducing the number of said ATTTA sequences and the number of said polyadenylation signal sequences in the coding sequence.

122. (New) A method of making a structural gene that encodes a protein, the method comprising:

(a) starting with a coding sequence that encodes a polypeptide and that contains a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table II;

(b) reducing the number of said ATTTA sequences or the number of said polyadenylation signal sequences in a portion of the coding sequence by substituting sense codons for codons in said portion; and

(c) making a structural gene that comprises said portion with the substitute codons and the reduced number of ATTTA or polyadenylation signal sequences, wherein the structural gene comprises a nucleotide sequence that encodes a protein.

123. (New) The method of claim 122, wherein step (b) comprises reducing the number of said ATTTA sequences and the number of said polyadenylation signal sequences in said portion.

124. (New) A method of making a structural gene that encodes a protein, the method comprising:

(a) starting with coding sequences, from one or more structural genes that encode a polypeptide and that contain a plurality of sequences selected from the group consisting of ATTTA sequences and polyadenylation signal sequences listed in Table II;

(b) reducing the number of said ATTTA sequences or the number of said polyadenylation signal sequences in the coding sequences by substituting sense codons for codons in the coding sequences; and

(c) making a structural gene that comprises the coding sequences with the codons substituted according to step (b) and characterized by the reduced number of ATTTA or polyadenylation signal sequences, wherein the structural gene comprises a nucleotide sequence that encodes a protein.

125. (New) The method of claim 124, wherein step (b) comprises reducing the number of said ATTTA sequences and the number of said polyadenylation signal sequences in said coding sequences.

126. (New) A method of making a structural gene that encodes a protein, the method comprising:

(a) starting with an amino acid sequence of an insecticidal protein; and

(b) making a structural gene that comprises a coding sequence that encodes the amino acid sequence and that is devoid or substantially devoid of ATTTA sequences, or devoid or substantially devoid of polyadenylation signal sequences listed in Table II.

127. (New) The method of claim 126, wherein step (b) comprises making a structural gene that comprises a coding sequence that is devoid or substantially devoid of the ATTTA sequences and devoid or substantially devoid of the polyadenylation signal sequences.

128. (New) A method of making a structural gene that encodes a protein, the method comprising:

(a) starting with coding sequences that encode portions of one or more polypeptides; and

(b) combining the coding sequences to form a structural gene that encodes a protein,

wherein said coding sequences and the structural gene are devoid or substantially devoid of polyadenylation signal sequences listed in Table II.

129. (New) The method of claim 128, wherein said coding sequences and the structural gene are devoid or substantially devoid of ATTTA sequences.

130. (New) The method according to any one of claims 120-129, further comprising attaching a plant promoter to the structural gene.

131. (New) The method according to any one of claims 120-129, further comprising including in the structural gene a sequence that encodes an amino-terminal chloroplast transit peptide or a secretory signal sequence.

132. (New) The method according to any one of claims 120-129, further comprising attaching to the structural gene a 3' non-translated nucleotide sequence that comprises a plant polyadenylation signal.

133. (New) The method according to any one of claims 120-125, wherein the structural gene made according to the method is more highly expressed in a dicot plant cell than a structural gene that comprises the starting coding sequence(s) of step (a).

134. (New) The method according to any one of claims 120-129, wherein the structural gene made according to the method contains no ATTTA sequences.

135. (New) The method according to any one of claims 120-129, wherein the structural gene made according to the method contains no polyadenylation signal sequences listed in Table II.

136. (New) The method according to any one of claims 120-125, wherein the starting coding sequence(s) of step (a) has (have) an (A + T) content of about 62%.

137. (New) The method according to any one of claims 120-127, comprising avoiding the introduction of sense codons that are rarely found in plant genomes into the resultant structural gene.

138. (New) The method according to any one of claims 120-127, comprising avoiding, in the resultant structural gene, the introduction of sense codons that contain a TA doublet.

139. (New) The method according to any one of claims 120-127, comprising avoiding, in the resultant structural gene, the introduction of sense codons that contain a CG doublet.

140. (New) The method according to any one of claims 120-125, further comprising reducing the number of regions in the coding sequence(s) with greater than five consecutive adenine and thymine (A+T) nucleotides by substituting sense codons for codons in the coding sequence(s).

141. (New) The method according to any one of claims 120-129, wherein the structural gene comprises a coding sequence that does not contain more than five consecutive adenine and thymine (A+T) nucleotides.